D1

An example of such a biosynthetic pathway enzyme is ribulose-1,5- bisphosphate carboxylase-oxygenase ("Rubisco"), which is the enzyme in plants, green algae (including marine algae), and photosynthetic bacteria involved in fixing atmospheric carbon dioxide into reduced sugars. Rubisco is a true bifunctional enzyme; it catalyzes (i) carboxylation of ribulose bisphosphate ("RuBP") to form two molecules of 3-phosphoglycerate, and (ii) oxygenation of rubp to form one molecule of 3-phosphoglycerate and one molecule of 2-phosphoglycerate, at the same active site. The oxygenation reaction catalyzed by Rubisco (also called photorespiration) is a "wasteful" process, since it significantly reduces the amount of carbon fixed. Both CO_2 and O_2 compete for the same active site, although the Km for CO_2 is about an order of magnitude less than for O_2. In plants, as the temperature rises during the course of the day, photorespiration catalyzed by Rubisco increases relative to carbon fixation, reducing the energy efficiency of carbon fixation. This is because the solubility of CO_2 decreases with increasing temperature relative to O_2. During the course of evolution, Rubisco has been selected for carboxylation specificity (carboxylation specificity factor defined as the ratio of maximal velocity of carboxylation x Km for O_2 to maximal velocity of oxygenation x Km for CO_2). This specificity has evolved from about 10 in bacteria, to 50 in cyanobacteria, and to about 80 in higher plants. In photosynthetic bacteria and dinoflagelates, Rubisco is present as a dimer of a large subunit (Form II, L_2), and no small subunit is present. In cyanobacteria, green algae, and higher plants (C3 and C4 plants), Rubisco is present as multimeric (e.g., hexadecimeric) protein composed of two subunits, the large (L) subunit which is catalytic, and the small (S) subunit which is regulatory, formed into an enzymatically active multimer (e.g., L_8S_8 hexadecimer). Coding sequences for L and S subunits for various species are disclosed in the literature and Genbank, among other public sources, and may be obtained by cloning, PCR, or from deposited materials.

At page 37, lines 4-8:

D2

For illustration and not to limit the invention, examples of a desired Rubisco enzymatic phenotype can include increased RuBP carboxylase rate, decreased RuBP oxygenase rate, increased Km for O_2, decreased Km for CO_2, decreased ratio of Km for CO_2 to Km for O_2,